Physiology. — *"Contributions to the Study of serumanaphylaxis"* (1st communication). By J. G. SLEESWIJK, Foreign Member of the Pasteur Institute at Brussels. (Communicated by Prof. SPRONCK).

(Communicated in the meeting of January 30, 1909).

Of late the problem of anaphylaxis has attracted the particular attention of more than one investigator of immunity. On the one side the purpose is to answer the purely scientific question, how hypersensibility has to be explained, which in an organism may appear with respect to very different albuminous substances after such a material in some way or other has formerly been assimilated by the organism in question. But on the other hand the practical serumtherapy wishes to be delivered from the difficulties of the serumdisease, and tries to find means of preventing the dangers which, already with the first injection, but still oftener with an injection that is repeated not too soon, threaten the patient.

In the meantime the sphere of investigation has been examined in many a direction, the literature is increasing, but theory has still too frequently to complete what is wanting in a useful supply of facts. Therefore an extension of the latter is very desirable, if new points of view offer themselves there. This communication is to furnish a contribution to this. It contains in a few words some results of the first part of an investigation which was made in the Pasteur Institute at Brussels and which had the phenomenon of the serumanaphylaxis for its subject.

The literature will only be referred to, as far as this is strictly necessary to elucidate my explanation [1]).

It was THEOBALD SMITH who had observed that guinea-pigs which had served for the titration of diphtheria-serum, and which accordingly had been treated previously with small quantities of diphtheriatoxine and antitoxic horse-serum, after a certain period of incubation had become extremely sensitive to a second injection of horse-serum, that they reacted thereupon as upon the administration of a strong poison and — in proportion to the dose — very often perished. OTTO proved that with nothing but horse-serum (without toxine) this hypersensitiveness was also obtained, whilst ROSENAU and ANDERSON proved that also with the aid of other sera such an anaphylactic state could be called into life, and that for each serum in a specific

[1]) For an ampler discussion about the present state of the problem I beg to refer to a critical study from my own hand, which is shortly to appear in the "Zeitschrift für Immunitätsforschung und experimentelle Therapie".

sense. Since that time the guinea-pig has become the fit test-animal for such investigations.

I have provisionally confined myself to the study of horse-serum, also because the knowledge of this in connection with the origin of our therapeutic sera has the most practical importance.

While a normal guinea-pig bears an intraperitoneal or subcutaneous injection of 5 cm^3 horse-serum without any perceptible symptom of disease, such an animal (of 250 to 300 grammes) mostly perishes however under typical symptoms of intoxication, when about 12 days before it has been treated with a small dose of the same serum (e.g. $\pm$ $^1/_{100}$ cm^3.). Instead of immunity (*pro*phylaxis), which usually follows on the administration of a larger dose, here a state of hypersusceptibility or *ana*phylaxis (Richet) has arisen. The horse-serum completely harmless in itself, plays in this case for the sensitized guinea-pig the part of a heavy poison. The first sensitizing injection must therefore have caused such changes in the organism as to change the second serum-injection into a toxic one.

This process of reaction no doubt belongs to the symptoms of immunity, and consequently it ought to be studied with the aid of the methods that the doctrine of immunity has procured. It was therefore a matter of course that the question was asked: Is in the process in question alexine fixed?

Otto[1]) answers this question in the negative, in my opinion wrongly. For repeated observation taught me that a sensitized guinea-pig, which reacts upon the second serum-administration with symptoms of intoxication some time after that injection produces a serum that is exceedingly poor in haemolytic alexine (sensitized red corpuscles serving as test-object). A short time (5—10 min.) after the toxic injection the alexic power of the pig-serum is still the same; after this it decreases gradually and rather rapidly, so that after $^1/_2$—1 hour it has become minimum. In this period the animal mostly dies. If it recovers, however, the alexine is also seen to increase again, so that $1^1/_2$—2 hours after the injection it has returned again to the normal level or even higher. This course of things might be graphically represented by means of a curve. In a normal, not anaphylactic guinea-pig the alexine-quantity of the serum remains constant under the same experimental circumstances.

Now, if the blood is not examined at the right moment, or not at several moments during the stage of intoxication, the chances are that one is too early (when the alexine has not yet disappeared) or

[1]) Münch. med. Woch. 1907, no. 34.

also too late (when it has recovered itself again). I presume that OTTO has thus been led astray.

I have still to add here that, if the second toxic injection is applied not in the abdomen or subcutaneously, but in the circulation — in consequence of which the symptoms of intoxication show themselves very soon and pass very quickly — these symptoms may already be present even before the alexine has disappeared from the serum of the animal. From this may be concluded that the symptoms of poisoning are not the consequence of the loss of alexine, but that these two are processes running parallel, independent of each other, but both having a common cause. And this can be no other but the reciprocal influence of the horse-serum administered (the antigen) and the reaction-products, specific for this, of the sensitized organism, arisen after and in consequence of the first injection of the alien serum.

This being settled, we continue asking ourselves: where are these reaction-products to be found — probably a particular kind of anti-bodies? Where do we meet with such materials as show a particular and specific affinity to horse-serum?

In order to answer this question, of course the first thing done was to examine the serum of sensitized guinea-pigs, but without any special result. For in not a single combination such serum gives a precipitate with horse-serum. Another possibility for the disappearance of the alexine from the serum of the intoxicated animals might still be found in the presence in their circulation of antialbuminoid sensibilisators of GENGOU. But also these seem to be wanting; I have repeatedly been able to convince myself that anaphylactic serum, again in not a single combination with horse serum, is able to fix alexine. On the other hand I have been able to prove that the serum of the sensitized pigs reacts antialexically with respect to fresh horse-serum, and especially during the stage in which after the toxic injection the original alexine has disappeared from the circulation. Although I now reserve to myself the duty to revert to the meaning of this fact on a future occasion, yet it seems to me that this formation of antialexines (which we also meet with at the usual serum-immunity) does not bring us much nearer to the explanation of just the anaphylactic complex of symptoms.

But if not the fluids, it is perhaps the cells that can bring us a step onward? — I have applied to the erythrocytes of the guinea-pig, and it has appeared to me that washed normal pig-blood, brought in contact with horse-serum, whilst a sufficient quantity of physiological solution of sodiumchloride is present, is able to fix

from the serum the substance that is toxic for sensitive animals.[1])

This procedure was already for another reason known in the immunity-literature, because in this way also the alexine from the horse-serum is fixed upon the blood of the guinea-pig.[2])

Serum treated thus has for anaphylactic animals lost its poisonousness, and this fact seems to me to open a new point of view. For it proves that there exists affinity between the toxic principle of horse-serum and cellular elements already of the normal pig-organism. The supposition does not seem to be too bold that also other elements of tissue or organs of the guinea-pig are subjectable to such a fixation, and that this affinity is still enhanced in the anaphylactized animal. The reaction between the horse-serum and the sensitive elements — especially those of the central nervous system — would then give rise to the action of the anaphylactic shock, whilst by the side of this the secondary fixation of the alexine would be the consequence of this reaction to be observed in the serum. Starting from these facts and considerations I continue my investigation in this direction.

In the meantime it is worth while to point out here that already some time ago v. BEHRING drew the attention to the paradoxical fact that a horse containing abundant diphtheria-antitoxines in its blood, can yet react upon a relatively small dose of toxine with symptoms of poisoning and even with death. Therefore v. BEHRING presumed the existence of an *histogenetic hypersensibility*, which hypothesis, in connection with what precedes, grows more probable.

To the many attemps made by different investigators with varying results, to deprive horse-serum of its toxicity by the help of physical or chemical means, I have tried to add another, which had a satisfactory result. I have namely submitted to dialysis horse-serum in so-called "Fischblasencondome". From this it appeared that the arising precipitation, dissolved in a physiological salt-solution, shows no trace of toxicity with respect to sensitized guinea-pigs, whilst the serum floating on the surface and free from salt (before the animal-experiment reduced to isotonical proportions), gradually loses its poisonousness during the process of dialysis.

Now the proof for the non-poisonousness just of the filtrate is not devoid of importance, because former investigations have shown that in dialysing antitoxical horse-serum the diphtheria-antitoxines (which

[1]) Take for 1 vol. serum: $1^1/_2$ vol. blood and 2 vol. salt solution. — On simple dilution with salt-solution in the same proportions, the serum retains its toxicity.

[2]) See about the meaning of this phenomenon: Ehrlich and Sachs, Berl. Klin. Woch. 1902, no. 21 and Bordet and Gay, Annales Pasteur 1906.

are fixed to the soluble globulines) are to be found back quantitatively in the filtrate. Thus the way has been paved to obtain an antitoxical solution, at the same time free from anaphylactic by-actions, — which might be of great use to the serum-therapy. Erelong I hope to be able to give further information about this subject.

Mathematics. — "*The types of bilinear* ∞^1*-complexes of* M_{r-2}^n *in* Sp_r." By Mr. LUCIEN GODEAUX, at *Liège*. (Communicated by Prof. SCHOUTE).

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I have been recently [1]) investigating which were the essential characteristics of the most general type of the bilinear complex of conics in Sp_3; it is now my purpose to extend my work to the linear space Sp_r with r dimensions.

Let there be ∞^r varieties M_{r-2}^n with $r-2$ dimensions and of order n. Any one of these varieties is entirely situated in a linear space Sp_{r-1} of the fundamental space Sp_r. Let us say that these ∞^r varieties form a ∞^1-complex.

The characteristics of such a complex are:

1. The number μ of the M_{r-2}^n situated in a general Sp_{r-1} of Sp_r.

2. The number ν of the M_{r-2}^n passing through a fixed point and the Sp_{r-1} of which passes through a Sp_{r-2} containing the chosen fixed point.

The aim we have here in view is the determination of the essential properties of the most general ∞_1-complex L having the characteristics $\mu = 1$, $\nu = 1$.

Let us notice that all the varieties M_{r-2}^n of Sp_r are the sections by the Sp_{r-1} of the varieties V_{r-1}^n with $r-1$ dimensions and of order n of a linear system $\binom{n+r-1}{n}-1$-times infinite K.

The M_{r-2}^n of L are evidently situated on the V_{r-1}^n of an ∞^r-system K' contained in K.

[1]) *Détermination des variétés de complexes bilineaires de coniques*. Bull. de l'Acad. Roy. de Belgique 1908.